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THE pH OF FRESH AND STORAGE EGGS

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BULLETIN NO. 335

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The pH of Fresh and Storage Eggs*

By S. E. ERIKSON, R. E. BOYDEN, J. HOLMES MARTIN and
W. M. INSKO, JR.

INTRODUCTION

A study of some of the constituents of hen's eggs has been undertaken for the purpose of investigating certain factors that may influence their nutritive quality. The study includes fresh and storage eggs and deals with the determination of the hydrogen ion concentration of both yolks and whites, and the amounts of copper, iron, calcium, phosphorus, lecithin, and cholesterol in the yolk together with hatchability, fertility and production studies. The conditions under which the hens were kept during the production of the eggs differed as to sunshine, cod liver oil and bluegrass range. The section of the study reported here deals with the pH of the yolks and whites of fresh and storage eggs. Other sections will follow as the work is completed.

From the experiments that have been reported, it appears that some constituents in the egg can be affected by variations in the diet of the hen while others cannot. Bethke, Kennard and Sassaman(1) for instance, have shown that feeding cod liver oil to laying hens effectively increases the vitamin D potency of the eggs. Studies in this laboratory (unpublished) show that the amount of vitamin D in the egg yolk varies with the administration of cod liver oil and sunshine. On the other hand, Elvehjem, Kemmerer, Hart and Halpin(2) added iron and iron plus copper, as sulfates, to the ration of hens but found no increase

*A common way of expressing hydrogen ion concentration is as pH. This is the logarithm of the reciprocal of the hydrogen ion concentration. A neutral solution is pH 7. Numbers larger than 7 indicate alkalinity; those smaller, acidity. The smaller the number, the more acid the solution.

in the iron or copper content of eggs. Cunningham,(3) also, found that copper added to the diet of the hen affected neither the copper nor the iron content of the eggs. Buckner, Martin, and Peter(4) found that the presence or absence of oystershell in the diet of the hen had no appreciable effect on the calcium content of the liquid portions of the eggs. Hughes, Payne and Latshaw(5) found that the administration of ultra-violet light ten minutes daily to confined pullets increased the average weight of calcium in the egg shell and the percent of calcium and phosphorus in the white and yolk of the eggs. Hart and co-workers(6) found that irradiation of hens with ultra-violet light markedly increased the amount of lime in the shell altho the calcium and phosphorus content of the whites and yolks was not distinctly different from those of non-irradiated hens.

In order, therefore, to get a more complete understanding than is at present available of certain constituents that may or may not affect the nutritive qualities of hens' eggs and their fertility and hatchability, a general procedure for the problem was established which is described in the following section.

GENERAL PROCEDURE

One hundred and fifty Single Comb White Leghorn pullets of approximately the same age, size and sexual maturity were divided into 6 pens of 25 each and placed in houses of identical construction and orientation. All the pullets were from high-laying parent stock in the Kentucky Experiment Station breeding flock. The pens were divided into 3 pairs, and one of each pair received cod liver oil in addition to the following all-mash which was fed *ad libitum* to all pens:

All-mash mixture fed dry in hoppers:

Ground yellow corn	650 pounds
Wheat bran	100 pounds
Wheat middlings	100 pounds
Meat scrap (50% protein).....	75 pounds
Dried buttermilk	25 pounds
Finely ground limestone	20 pounds
Steamed bone meal	20 pounds
Salt	10 pounds

Coarsely ground limestone (approximately 99% CaCO_3) and water were also available to all pens at all times. The pens were treated as follows:

- | | |
|--------------|--|
| First pair. | Confined to house, no direct sunshine, only sunshine thru common window glass.
Pen 1. No cod liver oil. Pen. 2. Cod liver oil, 2 percent of the mash. |
| Second pair. | Confined to house and wire-screened, wire-floored sun porch.
Pen. 3. No cod liver oil. Pen 4. Cod liver oil, 2 percent of the mash. |
| Third pair. | Allowed range of bluegrass yard.
Pen 5. No cod liver oil. Pen 6. Cod liver oil, 2 percent of the mash. |

Two males were placed in each pen a week before beginning collecting eggs for incubation, and were replaced if during the hatching season they showed sign of loss of vitality.

Eggs from six hens in each pen were collected for the chemical analyses on every other ten-day period, from February 19, 1930, to May 29, 1930. Hens were chosen whose previous egg production record showed an average of 5 or 6 eggs in 10 days. The eggs from these same hens were collected on the alternate ten-day periods to determine fertility and hatchability. All hens were trapnested and the eggs were collected daily at 7:30 and 10:30 A. M. and at 1:00 and 3:45 P. M.

To test the effect of storage, the eggs that were collected from May 29 to June 30, 1930, were placed in cold storage each evening. A temperature between 30° and 33° Fahrenheit was maintained until February, when the eggs were removed to the laboratory for one-half day to sort them from other experimental eggs laid at the same time. They were then placed in another cold storage room where a temperature of between 30° and 38° Fahrenheit was maintained until they were taken out for analysis during the summer of 1931.

The first investigation to be reported in this series deals with the hydrogen ion concentration of the yolks and whites of fresh and storage eggs. A preliminary discussion of methods is taken up first.

Part 1. Procedure for Determining Hydrogen Ion Concentration of Eggs

In preparing to investigate the pH of eggs, it was necessary to decide upon a method that would assure the greatest possible accuracy at the least possible expenditure of time, because the project planned involved the analysis of a fairly large number of eggs every day over a period of five or six months.

Work reported by other investigators on eggs indicates that a loss of carbon dioxide occurs, beginning very shortly after the egg is laid. It is necessary, therefore, to take this fact into consideration in deciding upon a method. Healy and Peter(7) used a colorimetric method. Sharp and Powell(8) used the electrometric method with a hydrogen electrode, and Baird and Prentice(9) used the electrometric method with a quinhydrone electrode. All these found that the pH of the white rises rapidly with increasing age. Considering the differences in the handling of the eggs, the results show a surprising agreement. Table 1 gives a comparison of the results obtained by the different methods. It will be noted that Sharp and Powell report a range of pH from 7.6 for the white of eggs a few hours old to 9.4 for eggs several weeks old. Baird and Prentice found that the white of eggs stored at 18°C, from 2 hours to 57 days, ranged in pH from 7.63 to 9.06. Healy and Peter give their results as pH 8.25 for eggs a few hours old to pH 9.4 for eggs 96 hours old. The pH of the yolk was found to be more stable than that of the white. Sharp(10) gives for yolk a range of pH 6.0 to pH 6.8. He explains this difference as due to passage of carbon dioxide from the yolk into the white.

It appears from Table 1 that while the initial loss of carbon dioxide is rapid, the increase in pH becomes progressively slower. This accords also with the results of Sharp and Whittaker(11) who report that in 24 hours the pH of the white may change as much as 1.0 pH unit when held at room temperature and that in six days it may change only as much as 0.8 pH unit more. The greater part of the change obviously occurred the first day.

Sharp and Powell(8) employed the technique of breaking the egg, pipetting it into a test tube and covering it with oil in order to reduce to a minimum the loss of carbon dioxide. Even with this procedure they felt that a pH of 7.6 is undoubtedly too high for fresh whites.

Since the consensus of opinion seems to be that differences in pH are caused by the loss of carbon dioxide, it was necessary for the problem being undertaken in this laboratory to have more specific information as to results obtained by the different methods on the same egg; also, as to whether a direct relationship exists between the loss of carbon dioxide and the time elapsing before the determination is made. The effect of dilution with distilled water was noted at the same time because it was felt that the process of mixing the egg with the water might hasten the release of the gas.

In trying out the colorimetric method Clark and Loeb buffers were made up for a range of pH from 6.0 to 9.8. It was found, however, that it was difficult for the workers in this laboratory to be sure of the color obtained with the yolk and that it was also difficult to get the indicator to mix thoroly and quickly with the white in a test tube. Placing a drop or two on a porcelain test tablet and mixing the indicator in it with a stirring rod gave even less satisfactory results.

The use of the hydrogen electrode offered the difficulty of saturating the material with hydrogen. Both the white and the yolk are so viscous that it takes a considerable length of time to saturate them. A bubbling hydrogen electrode could not be used because, obviously, dissolved carbon dioxide would be driven off. The Bailey hydrogen electrode, however, is tightly stoppered while the material to be tested is being saturated with hydrogen and, therefore, it is assumed that no loss of gas occurs at that time. With the quinhydrone electrode the possibility was present of both salt and buffer interference. It was decided, therefore, to check the results obtained with the two electrodes, using the same egg. For the quinhydrone, a saturated calomel cell and a gold electrode were used with a Leeds and Northrup

quinhydrone pH indicator. A motor-driven glass stirring rod mixed the contents of the beaker with the least possible amount of surface agitation. When making the determinations, the points of the indicator became constant in from 1.0 to 1.5 seconds, at which time the reading was taken. The accuracy of the electrodes and indicator were checked daily against a standard acetate buffer solution of pH 4.63 at 25°C.

For the hydrogen electrode a Leeds and Northrup potentiometer was used, the Portable d'Arsonval galvanometer and the Eppley Standard Cadmium Cell. For all the determinations distilled water was obtained from a Stokes Automatic Water Still, the condenser tube of which is lined with quartz.

It will be noted from Table 2 that the quinhydrone and the hydrogen electrode, up to a pH of 9.0, check each other to within less than a tenth of a unit. The greatest difference in values below 9.0 was 0.13 pH, the average being 0.03. Above 9.0 the greatest difference was 0.33 pH and the average was 0.08 pH.

From Table 3 it may be seen that standing for 50 minutes after the egg is opened seems to have no appreciable effect on the undiluted egg yolk. The average pH before standing was 6.18 and that after standing was 6.16. Standing for 30 minutes likewise had practically no effect on the undiluted egg white, the average values being 9.13 before standing and 9.14 after standing. The difference in the length of time for standing of yolk and white was an inadvertent one due to the difficulty experienced in saturating the undiluted egg yolk with hydrogen.

Yolk and white diluted 1:1 with distilled water seemed not to be affected by being allowed to stand open 20 minutes before the determination was made. Table 4 shows that the average figure for yolk was 6.22 of determinations made at once and 6.21 of determinations made after the diluted yolk had stood 20 minutes. The average of the determinations made on diluted white when the egg was first opened was 9.22 as compared to an average of 9.23 for determinations made after the white had stood 20 minutes.

The effect of allowing the opened egg to stand then diluting

it 1:1 with distilled water and making the determination at once was likewise negligible, shown in Table 5. Not enough eggs were used to give averages but even on individual eggs, it is apparent that it makes little or no difference whether the dilution is made before or after the eggs are allowed to stand. For instance, 6.03 was found to be the pH of yolk before standing and 6.02 the pH after it had stood 30 minutes and was then diluted. The pH determined at once on egg yolk not diluted was 6.19. On the same egg allowed to stand 80 minutes and then diluted 1:1, it was 6.14. A pH of 6.12 was found for a yolk allowed to stand 30 minutes and then diluted, and the same value when the determination was made on the undiluted yolk. The differences found for the whites in this case were likewise not any greater than the variations between individual eggs.

When the comparisons were made between the eggs that had stood diluted and those undiluted on which the determinations had been made at once, Table 6, no greater differences were found than those between individual eggs, the average pH for the undiluted eggs that did not stand being 5.95 for the yolks and 9.05 for the whites, while the average pH for the eggs diluted and then allowed to stand was 6.02 for the yolks and 9.19 for the whites.

Eggs allowed to stand diluted and undiluted show slight differences, Table 7, the average values for the undiluted ones being 6.09 for the yolk and 8.94 for the white, while the diluted yolk showed an average of 6.20 and the diluted white an average of 9.07.

In taking the samples for the pH determinations on egg white, it is difficult to get a fair sample, that is, an equal amount of thick and thin white. Because the viscosity of colloidal substances seems to be affected by hydrogen ion concentration, it was thot necessary to determine whether the thick and thin white of the same egg have different pH values. Accordingly the two portions were separated, which can easily be done on fresh eggs by allowing the thin part to run off from the thick part, and the pH value found for each part. The results given in Table 8

show that practically no difference exists between the two portions. The average pH values for both thin and thick whites were the same, namely 8.42. This seems to indicate that the liquefaction of thick white to watery white, which occurs in storage eggs, is not due directly to the increased pH which Sharp,(10) Sharp and Powell,(12) and others have found in storage eggs.

From the results obtained in these comparisons, it was concluded that as a procedure to be adopted for the determination of the pH of eggs it would be safe to dilute the material 1:1 with distilled water, and that standing open as long as half an hour caused no appreciable difference in the values obtained. The results of the tests on standing seem to indicate that the loss of carbon dioxide occurs very quickly upon the breaking of the egg and that further loss is a slow one. The hydrogen ion concentration found has to be considered, therefore, as that of the egg, the gas pressure of which has been equilibrated with room air. It is conceded that this is probably not the accurate figure for the pH of yolk and white of egg but since, in the problem undertaken in this laboratory, the comparison between the pH of eggs produced under different conditions was the point to be emphasized, it was considered practical to disregard the swift initial loss of carbon dioxide which seems to occur as soon as the egg is broken and which, as a matter of fact, seems to start as soon as the egg is laid. To have carried precaution against loss of carbon dioxide to a logical conclusion would have meant receiving and breaking the egg under oil, a procedure which would have made it impossible, in the problem undertaken here, to get comparative data.

The quinhydrone electrode was adopted for fresh eggs, but because the pH values for the whites of storage eggs would probably run higher, the hydrogen electrode was decided upon for storage eggs.

After the work in Part 2 had been finished, the question arose as to whether the conclusion drawn in Part 1 was valid, namely, that the pH of eggs does not change greatly during the

first 30 minutes after they are opened except for the rapid initial loss of carbon dioxide that occurs as soon as the shell is broken. For instance, the question was asked, if eggs of a lower pH had been tested to determine the effect of length of time of standing open, would they have been less stable? Therefore, in order to know whether the conclusion was justified another series of determinations was made, the results of which are reported in Table 9. Freshly laid eggs were opened and allowed to stand 15, 30 and 45 minutes in order to compare the values obtained at the expiration of the three time limits with values obtained on the same sample when the pH determination was made as soon after the egg was opened as it was possible to make it. For the work reported in Part 2, eggs never stood open as long as 45 minutes before the determination was made. Twenty minutes was the outside limit but, in order to know what changes might occur, the 45 minute period was included also.

Inspection of Table 9 shows that standing open 30 minutes or less has little effect on the pH, the average values for egg white showing a difference of .03 pH for 15 minutes and .01 pH for 30 minutes. For yolks the difference was .02 pH for 15 minutes and .01 pH the difference for 30 minutes. Standing open for 45 minutes gave a difference of .07 pH for whites and .06 pH for yolks. From these results it is apparent that the length of time the eggs stood open before the determination was made did not affect the average results sufficiently to invalidate the conclusions drawn in Part 2.

It was held to be of interest, also, to tabulate the results of pH determinations given in Part 2 to show the effect of time of standing after the egg was collected until the determination was made. From Table 10 it is apparent that the eggs that stood from one-half hour up to nine hours show little consistent increase in pH in either yolks or whites. The length of time of standing after the collection was made does not, of course, indicate the true age of the egg, but in view of the fact that the collections were made at 7:30 A. M., 10:30 A. M., 1:00 P. M. and 3:45 P. M., and since the eggs brought to the laboratory at each

collection were all analysed before those of the next collection were analysed, it is evident that the time of making the analysis after the eggs were laid did not vary from period to period. From these figures it does not appear that differences in the pH of eggs averaged either per pen or per period could have been affected by variations in the length of time the eggs stood after they were laid until the determination was made.

Table 1. Comparison of pH of eggs as ascertained by different investigators.

Colorimetric Healy and Peter*		Quinhydrone electrode Baird and Prentice**		Hydrogen electrode Sharp and Powell†	
Age of egg	pH	Age of egg	pH	Age of egg	pH
Yolk					
New laid	6.2	2 hours	6.05	New laid	6.0
	6.6	8 hours	6.07	New laid	6.04
	6.4	18 hours	6.04	92 hours	6.04
48 hours	6.3	44 hours	{ 6.01	8-10 days	6.14
92 hours	-----		{ 6.04	20-22 days	6.2
96 hours	6.4	68 hours	6.05	36 days	6.46
8 days	6.2	98 hours	6.01		
10 days	-----	5 days	5.97		
14 days	6.4	7 days	5.99		
21 days	6.3	8 days	6.05		
28 days	6.6	15 days	6.05		
36 days	-----	22 days	6.09		
56 days	7.2	29 days	6.01		
		57 days	6.22		
White					
New laid	8.2	2 hours	7.63	New laid	7.60
	8.2	8 hours	8.21	24 hours	8.89
	8.4	18 hours	8.34	48 hours	9.05
24 hours	-----	44 hours	{ 8.54	72 hours	9.18
48 hours	9.0		{ 8.51	5 days	9.33
72 hours	-----	68 hours	8.76	7 days	9.39
96 hours	9.4	98 hours	8.83	10 days	9.37
8 days	9.5	5 days	8.93	14 days	9.46
10 days	-----	7 days	8.99	21 days	9.38
14 days	9.6	8 days	9.01	28 days	9.36
21 days	9.6	15 days	9.03	56 days	9.25
28 days	9.7	22 days	8.98		
56 days	9.4	29 days	9.02		
		57 days	9.06		

*Amer. Jour. Physiol. 74: 363; 1925. Table 3, p. 364.

**Analyst 55: 20, 1930. Part of results from Table 1, p. 21 and Table 2, p. 22.

†Indus. and Engin. Chem. 23: 196; 1931, Table 1. At 25° C.

Table 2. Comparison of the pH of egg yolk and egg white as determined by the hydrogen electrode and quinhydrone electrode.

Sample No.	YOLK		WHITE	
	H. Electrode	Quin. Electrode	H. Electrode	Quin. Electrode
101	6.09	6.19	9.07	9.02
106	6.19	6.14	9.27	8.94
102	6.04	6.17	9.19	9.33
107	6.06	6.07	9.21	9.13
103	6.18	6.12	-----	-----
108	6.06	6.12	-----	-----
104	-----	-----	9.34	9.16
105	-----	-----	8.92	8.97
Average	6.10	6.13	9.17	9.09

Table 3. Comparison of the pH of undiluted egg yolk and egg white after standing, with the yolk and white not allowed to stand.

Electrode	Sample No.	YOLK		Sample No.	WHITE	
		Detn. made at once	Detn. made after standing 50 min.		Detn. made at once	Detn. made after standing 30 min.
		pH	pH		pH	pH
Hydrogen	48	6.16	6.18	52	9.00	9.02
Hydrogen	49	6.18	6.17	53	8.93	9.08
Hydrogen	50	6.26	6.24	54	9.06	9.13
Hydrogen	51	6.29	6.26	55	9.10	9.10
Hydrogen	-----	-----	-----	56	9.19	9.23
Hydrogen	-----	-----	-----	57	9.25	9.23
Quin-	102	{ 6.04	6.06		9.19	9.21
hydrone		{ 6.17	6.07		9.33	9.13
Average		6.18	6.16		9.13	9.14

Table 4. Comparison of the pH of diluted egg yolk and egg white after standing, with diluted yolk and white not allowed to stand.

Electrode	Sample No.	YOLK		Sample No.	WHITE	
		Detn. made at once diluted 1:1	Allowed to stand 20 min. then diluted 1:1		Detn. made at once diluted 1:1	Allowed to stand 20 min. then diluted 1:1
		pH	pH		pH	pH
Hydrogen	20	6.48	6.49	25	9.29	9.26
Hydrogen	21	6.20	6.22	26	9.32	9.33
Hydrogen	22	6.18	6.17	27	9.31	9.31
Hydrogen	23	6.19	6.18	28	9.40	9.44
Hydrogen	24	6.23	6.19	29	9.39	9.37
Quinhydrone	100	6.03	6.02	100	8.61	8.65
Average		6.22	6.21		9.22	9.23

Table 5. The effect of dilution and of standing on the pH of egg yolk and egg white.*

Sample No.	Detn. made at once diluted 1:1	Detn. allowed to stand 30 min. then diluted 1:1	Detn. made at once (not diluted)	Detn. allowed to stand 80 min. then diluted 1:1	Detn. allowed to stand 65 min. (not diluted)	Detn. allowed to stand 30 min. (not diluted)
Yolk	pH	pH	pH	pH	pH	pH
300	6.03	6.02	-----	6.14	-----	-----
301	-----	-----	6.19	-----	-----	-----
302	-----	-----	6.17	-----	6.07	-----
303	-----	6.12	-----	-----	-----	6.12
White						
300	8.61	8.65	-----	-----	-----	-----
301	-----	-----	9.02	8.94	-----	-----
302	-----	-----	9.23	-----	9.13	-----
305	-----	8.92	-----	-----	-----	8.97

*Quinhydrone electrode.

Table 6. Comparison of the pH of diluted egg yolk and egg white after standing, with undiluted yolk and white not allowed to stand.

Electrode	Sample No.	YOLK		Sample No.	WHITE	
		Detn. made at once (not diluted)	Detn. made at end of 70-75 min. diluted 1:1		Detn. made at once (not diluted)	Detn. made at end of 20 min. diluted 1:1
		pH	pH		pH	pH
Hydrogen	37	5.81	5.95	30	9.01	9.22
Hydrogen	38	5.74	5.83	31	9.06	9.18
Hydrogen	39	6.07	6.18	32	8.93	9.14
Hydrogen	40	5.77	5.85	33	9.15	9.28
Hydrogen	-----	-----	-----	34	9.10	9.22
Hydrogen	-----	-----	-----	35	9.04	9.21
Hydrogen	-----	-----	-----	36	9.06	9.22
Quin-	101	{ 6.09	6.19		9.07	9.29
hydrone		{ 6.19	6.14		9.02	8.94
Average		5.95	6.02		9.05	9.19

Table 7. Comparison of the pH of diluted egg yolk and egg white, with the undiluted yolk and white after standing.

Electrode	Sample No.	YOLK		Sample No.	WHITE	
		Detn. made after standing 45-75 min. (not diluted)	Detn. made after standing 30-35 min. diluted 1:1		Detn. made after standing 30 min. (not diluted)	Detn. made after standing 20 min. diluted 1:1
		pH	pH		pH	pH
Hydrogen	41	5.87	5.90	45	8.86	8.95
Hydrogen	42	5.99	6.22	46	8.91	9.08
Hydrogen	43	6.11	6.35	47	8.85	8.99
Hydrogen	44	6.36	6.43	-----	-----	-----
Hydrogen	103	6.06	6.18	-----	-----	-----
Quin-	103	6.12	6.12	-----	-----	-----
hydrone						
Hydrogen	104	-----	-----	-----	9.12	9.34
Quin-	104	-----	-----	-----	-----	9.16
hydrone						
Quin-	105	-----	-----	-----	8.97	8.92
hydrone						
Average		6.09	6.20		8.94	9.07

Table 8. Comparison of the pH of the thin and thick portions of egg white.*

Sample No.	Thin white	Thick white
	pH	pH
305	8.24	8.29
306	8.53	8.44
307	8.51	8.57
308	8.41	8.63
309	8.41	8.27
310	8.17	8.09
311	8.57	8.67
312	8.50	8.42
Average	8.42	8.42

*Quinhydrone electrode.

Table 9. Effect on pH of fresh egg yolks and whites of standing open 15, 30 and 45 minutes.

Sample No.	YOLK		WHITE	
	Detn. made at once diluted 1:1	Detn. made at end of 15 min. diluted 1:1	Detn. made at once diluted 1:1	Detn. made at end of 15 min. diluted 1:1
	pH	pH	pH	pH
359	6.49	6.58	8.32	8.32
360	6.39	6.39	8.25	8.25
367	6.56	6.53	8.09	8.18
Average	6.48	6.50	8.22	8.25

Sample No.	Detn. made at once diluted 1:1	Detn. made at end of 30 min. diluted 1:1	Detn. made at once diluted 1:1	Detn. made at end of 30 min. diluted 1:1
352	6.58	6.59	8.41	8.45
357	6.56	6.46	7.35	7.34
365	6.56	6.64	8.21	8.21
Average	6.57	6.56	7.99	8.00

Sample No.	Detn. made at once diluted 1:1	Detn. made at end of 45 min. diluted 1:1	Detn. made at once diluted 1:1	Detn. made at end of 45 min. diluted 1:1
350	6.29	6.29	8.30	8.38
354	6.52	6.41	8.51	8.57
Average	6.41	6.35	8.40	8.47

Table 10. Effect on pH of fresh egg yolks and whites of standing from $\frac{1}{2}$ hr. to 9 hrs. after collection.

Hrs. after eggs were collected before detn. was made	No. obs.	Yolk pH	No. obs.	White pH
0.0	5	6.54	5	8.00
0.5	40	6.36	41	7.81
1.0	55	6.33	55	7.77
2.0	58	6.31	58	7.83
3.0	55	6.30	55	7.88
4.0	40	6.32	39	7.91
5.0	41	6.42	41	8.04
6.0	21	6.45	21	8.00
7.0	14	6.51	14	8.25
8.0	4	6.37	4	7.89
9.0	2	6.52	2	8.23

SUMMARY AND CONCLUSIONS

1. The quinhydrone and the Bailey hydrogen electrodes were compared for determining the pH of yolks and whites of eggs. They were found to check each other up to a pH of 9.0 to within one-tenth of a unit.

2. Standing open up to 30 minutes for egg white and 80 minutes for egg yolk did not seem to affect the pH.

3. A 1:1 dilution of egg yolk and egg white did not seem to affect the pH, whether the dilution was made before or after being allowed to stand.

4. No difference in pH was found between the thick and thin portions of the white.

5. The conclusion was drawn that for making comparisons between the pH of eggs produced under different conditions, the procedure could safely be adopted of diluting the yolks and whites 1:1 and determining the pH within 30 minutes after the eggs were broken. It was also concluded that the Bailey hydrogen electrode can be used for the determination of the pH of both fresh and storage eggs but that since the time necessary for making the determination is longer than when the quinhydrone electrode is used, the latter may be adopted for deter-

mining the pH of fresh eggs but not for the whites of storage eggs since the pH of these may go above 9.0.

6. After the work reported in Part 2 had been finished, a series of determinations was made on freshly laid eggs to determine the effect of standing open on eggs of a lower pH. The differences found were so small that it was felt the conclusion could still be drawn that standing open up to 30 minutes did not affect the pH.

7. From the tabulation made from Part 2 of the time of standing of eggs, it was concluded that standing, unbroken, up to nine hours was not a factor affecting averages of the pH values for the different pens and the different periods.

Part 2. The Hydrogen Ion Concentration of Yolk and White of Fresh and Storage Eggs

Very small variations of pH have such marked effect on cell metabolism that even slight differences may be expected to influence fertility and hatchability of eggs. On the other hand, cell contents are so highly buffered that in all probability only a profound influence could be expected to modify the initial hydrogen ion concentration of the egg as it is formed in the body of the hen. A review of the work that has been done on determination of the pH of eggs by different investigators has been given in Part 1 of this report. A comparison of their results shows a fair agreement of values, considering the variations in technique and methods used.

A preliminary study was made in order to obtain a more thorough understanding of the different factors that may affect the accuracy of the determination of pH. The study included observations on 535 eggs, the detailed results of which are on file at the Library of the Experiment Station of the University of Kentucky and some of the results of which are given in Part 1 of this report. From these results it was concluded that after what seems to be an initial and a very quick loss of carbon dioxide, neither dilution with distilled water nor standing open for 30 minutes, makes an appreciable difference in pH. Nor does it seem to make any difference whether the dilution is made when the egg is first opened or after it has stood 15 to 45 minutes. Also, in the preliminary tests, the thin and thick portions of the egg white were tested separately. No difference was found between the two portions of the same egg. Both the quinhydrone and hydrogen electrodes were used for the determinations. They checked each other up to a pH of 9.0.

In view of the results of the preliminary tests, therefore, the following procedure was adopted for fresh eggs: As soon as possible after the eggs were collected, the determination was made. The time varied from ten minutes up to six hours, and

in two instances it was as long as nine hours. The eggs were weighed, broken against the side of a beaker, the white dropped into one beaker and the yolk into another, from which weighed portions were taken for the determination of certain constituents. The chalaza and the yolk membrane were left in that part of the yolk on which the pH was determined. Both white and yolk were diluted with distilled water. The distilled water was obtained from a Stokes automatic water still, the condensing tube of which was lined with quartz. The quinhydrone gold electrode was used for the fresh eggs, with a saturated calomel cell. Agitation of the sample was done by means of an electric glass stirrer. The potentiometer used was a Leeds and Northrup hydrogen ion indicator. The accuracy of the electrode and indicator was checked daily against a standard acetate buffer solution which had a pH of 4.63 at 25° Centigrade.

The storage eggs were analyzed after they had been kept in storage for about a year. The number that could be analyzed in one day were taken out at one time. Each egg was weighed and a similar procedure to that for fresh eggs was followed except that a Bailey hydrogen electrode was used instead of the quinhydrone electrode, together with the usual Leeds and Northrup potentiometer, the Portable d'Arsonval galvanometer and the Eppley standard cadmium cell.

RESULTS AND DISCUSSION

The results of the determinations are shown as averages in Tables 11-20 and Figures 1-3.

Fresh Eggs. The averages of values for fresh eggs, shown in Tables 11 and 12, were made on each pen for each period and the mean was taken of the averages of the periods for each pen. Correlation coefficients between yolks and whites are shown in Table 12. Figure 1 illustrates the egg production and the relationship between the pH of yolks and whites.

It will be noted that there is very little variation between periods. In all the pens, the lowest pH average for whites is found in the third period which ran from March 28 to April 9.

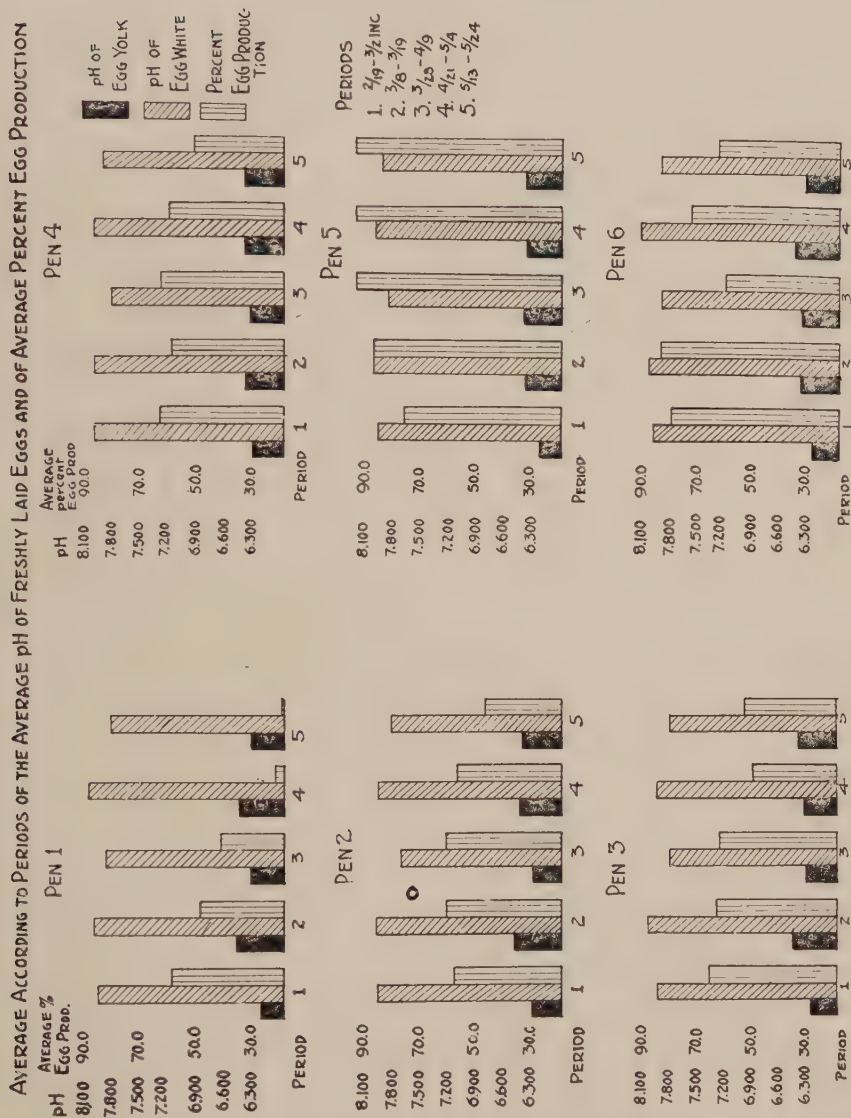


Figure 1. The pH of fresh eggs and average egg production of all pens.

Since the differences between individual eggs of the same hen and also between hen averages are often greater than the differences of the averages of the periods, it appears that a lower value for the third period, which occurs consistently in all the pens, has some significance even tho the differences between periods are small. If this is a logical deduction, then it is true for any other variation consistently occurring in all the pens even tho the differences are small. A similar variation is seen in the pH of the yolks, the averages of which are lowest in the first period in all pens, with an increase to the highest value in the second period in all pens, except 4 and 6, which show the highest value in the fourth period. Figure 2 shows that during the third period, the pH of the yolks, as well as that of the whites, is decreased. The averages per period of all the pens give two peaks, one for the second and one for the fourth period, with the low values in the first, third and fifth periods. Because these variations occur in all the pens, regardless of whether they have sunshine or cod liver oil or are confined without sunshine, it was thot either temperature or barometric pressure might be the influencing factor. Accordingly, the barometric pressure and lowest temperature of the days on which the eggs were laid were compared with the pH of the eggs. No relationship was observed. Nevertheless, it might be argued that if weather conditions have any influence they will be more apparent while the egg is being formed than at any time later. Therefore, averages of barometric pressure and temperatures for five days before the eggs were laid were checked and again no relation could be noted between the weather factors and the pH of the eggs. Neither the egg production of the hens from which eggs were collected for pH determinations nor that of the entire pens seemed to be affected by weather conditions during the periods over which eggs were collected. On only two days between February 19, 1930, and May 24, 1930, did the temperature drop below 20° F., reaching its low point of 13° F. on March 3, 1930. The warmest day between these dates of collection was on May 6, 1930, the outside temperature reaching 88° F.

The effect on the pH of yolks of fresh eggs of giving the hens cod liver oil is slight. It will be noted in Figure 3 and Table 13 that it seems, however, to be consistent; that is, the average pH of pen 2 is greater than that of pen 1; of pen 4 is greater than of pen 3, and of pen 6 is greater than of pen 5. Nevertheless, it is doubtful whether the differences are real. The averages given in Table 13 are the means of the averages for each

HYDROGEN ION CONCENTRATION OF WHITES & YOLKS OF FRESH AND STORAGE EGGS

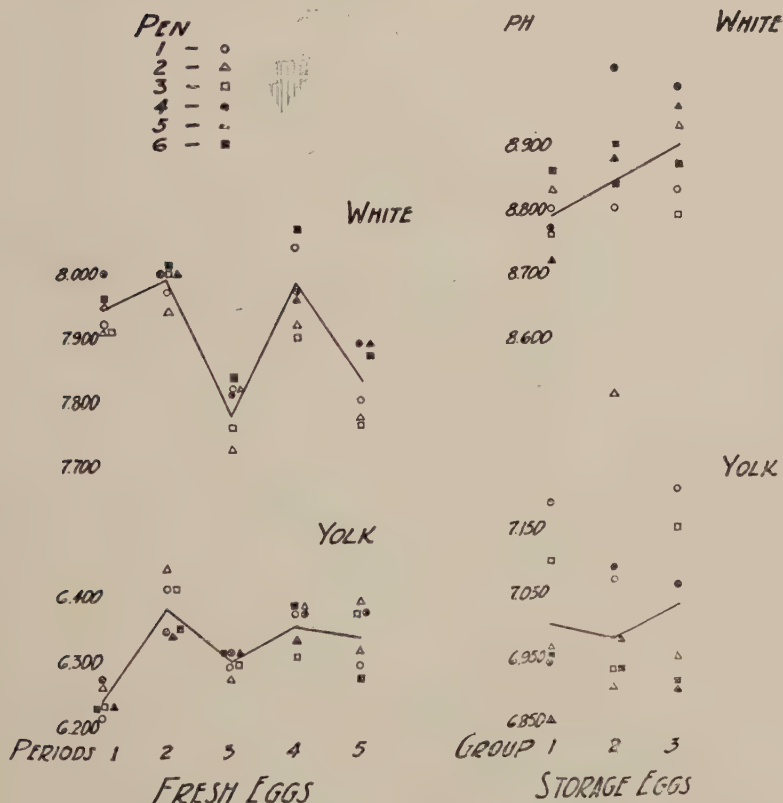


Figure 2. Average by periods of hydrogen ion concentration of white and yolk of fresh and storage eggs.

period and are, therefore, slightly different from the averages given in Tables 11 and 12, which are the means of all the eggs in each pen for all the periods. In the latter tables, the value for

HYDROGEN ION CONCENTRATION OF WHITES & YOLKS OF FRESH & STORAGE EGGS

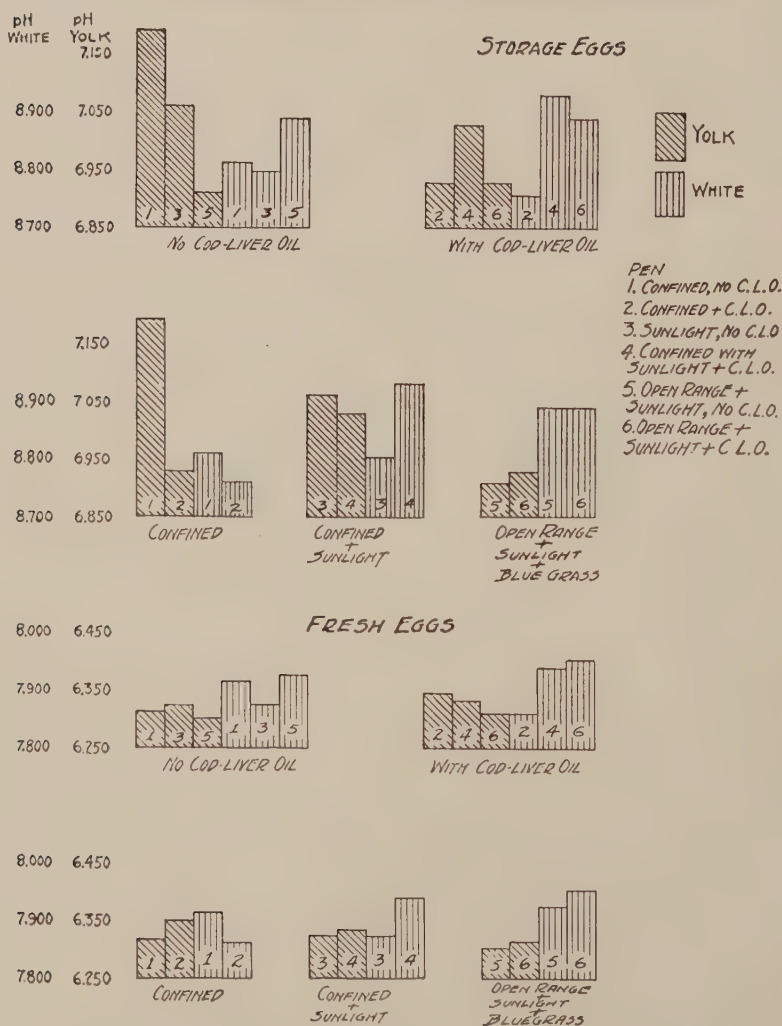


Figure 3. Pen averages of hydrogen ion concentration of white and yolk of fresh and storage eggs.

pen 3 is 6.317 and for pen 4 is 6.313 for yolk, and for white it is 7.894 for pen 3 and 7.915 for pen 4. Since the differences are so small, it is scarcely possible to consider them significant. This is borne out by the probable errors of the differences of the means given in Tables 14 and 15. According to these figures, none of the differences are significant. Nevertheless, it is interesting to note that when the average of averages is taken, the effect of cod liver oil seems to be consistent, even tho it is slight. The fact, also, that the lowest number of observations made on any pen was 114 for pen 1 and that the others were 153, 154, 143, 199 and 209, gives added weight to consistent variations.

The effect of sunshine and of bluegrass range appears to be negligible. Figure 3 and Table 13, show no consistent differences between pens 1, 3 and 5, getting sunshine thru window glass, sunshine on a screened porch and bluegrass range, all without cod liver oil, nor are any consistent differences to be noted in the corresponding pens given cod liver oil. Again the probable errors of the differences of the means indicate that whatever differences are found are of no significance.

Storage Eggs. Since one egg yolk does not contain enough material for the analysis of all the constituents chosen for study, the eggs laid by each hen were divided into three equal groups for each hen and these were designated Groups 1, 2 and 3. Lecithin and cholesterol were determined in the eggs of Group 1, calcium and phosphorus in those of Group 2 and copper and iron in those of Group 3. Hydrogen ion concentration was determined in all the eggs of each group but the calculations were made by groups in order to facilitate future correlations between pH and the constituents noted above. These correlations will be reported in a later paper.

Tables 16 and 17 give the averages of the pH values per group for each hen and also the average value for the pen. The values for the pen averages are the means of all the observations for each pen, while the pen averages in Table 18 are obtained by finding the mean of the pH of the eggs from each hen and then taking the mean of these values. In noting the averages per hen

per group in Tables 16 and 17 and comparing them with the pen averages, the fact that stands out is that wider variations are found in the hen averages per group than between the averages for the different pens. For instance, disregarding single determinations, the largest difference in mean pH of the yolk, Table 16, is from 6.61 for hen 391 in pen 1, to 7.87 for hen 386 in the same pen; and for white, Table 17, from 8.40 for hen 421 in pen 2, to 9.10 for hen 410 in the same pen. Differences between pen averages are noticeably smaller, the widest being 0.118 pH unit for yolk, between the values, 6.900 for pen 5 and 7.018 for pen 3, and 0.150 pH unit for white, between the values, 8.810 for pen 2 and 8.960 for pen 4.

These data indicate that the different treatment given the six pens had little or no effect on the pen average. Statistically real differences, as may be noted in Table 20, are found between the pH of the white of pens 3 and 4, 2 and 4, and 2 and 6. Statistically real differences are also found between the pH of the yolks of pens 3 and 5 and of 4 and 6, as may be seen in Table 19. Because the differences do not occur consistently, they cannot be ascribed to the variables in the treatment of the different pens. It seems probable that the conditions of handling during storage may be responsible for whatever variations occurred. It is possible that there would have been no differences if the storage period had not been interrupted and the eggs changed from one place of storage to another, assuming that all the eggs could have been removed to the laboratory and analyzed at one time.

An equilibrium of carbon dioxide in the atmosphere surrounding the eggs may have been reached, according to Sharp and Powell(12) who say that when eggs are kept in a confined space, the white gives off carbon dioxide until equilibrium between the carbon dioxide of the air and that of the egg white is reached. It is possible that an equilibrium of pH would then also occur but since only the eggs to be analyzed each day were taken out at one time, repeated and successive disturbances of the surrounding atmosphere could not be prevented, with consequent variations in the pH of the eggs.

When the fresh and storage eggs are compared, it is noted that the pH of storage eggs, both yolk and white, is higher than that of fresh eggs. Taking the mean of the averages of the six pens, the value for storage egg white is 8.895 as compared with that of fresh eggs, 7.920. The mean value for yolks of storage eggs is found to be 6.976 as compared with fresh eggs, the mean value for which is 6.320. Storage, therefore, definitely increases the alkalinity of both yolk and white of eggs. However, the degree of alkalinity in storage eggs probably depends on the conditions under which they are kept, because the popular idea seems to be that storage eggs are more definitely alkaline than the ones tested in these experiments proved to be.

Correlations. The coefficients of correlation between the pH of the yolk and white are given in Tables 12 and 17. Very little correlation is shown between the yolk and white of the fresh eggs, which seems to indicate that the two substances are quite independent and rather thoroly separated as to ions by the yolk membrane. More correlation is found in the storage eggs, especially in pens 2, 5 and 6. Apparently the yolk membrane has become more permeable to ions or else the pressure of ions has become greater. Sharp (10) suggests that with increasing age there is a gradual passage of carbon dioxide from the yolk to the white and from the white to the outside of the egg. No suggestion has been made that an exchange of ions occurs between the yolk and the white.

Table 11. The pH of the yolk of fresh eggs.

Pen	Hen	Period 1 2/19-3/2	Period 2 3/8-3/19	Period 3 3/28-4/9	Period 4 4/21-5/4	Period 5 5/13-5/24	Av. pH per pen	Standard Deviation
1	386	6.206	6.376	6.386	6.523	6.278		
	387		6.467	6.197	6.178	6.164		
	391	6.229	6.374	6.248	6.405	6.372		
	392	6.262	6.473	6.242				
	395	6.190	6.458					
	402	6.164	6.321	6.393	6.366	6.351		
	Av.	6.210	6.412	6.293	6.368	6.291	6.324±.012	.197±.003
2	410	6.267	6.332	6.282	6.364			
	411	6.310	6.479	6.156	6.390	6.373		
	421	6.309	6.568	6.286	6.351			
	426	6.213	6.439	6.296	6.396	6.476		
	427		6.355	6.318	6.402	6.318		
	428	6.206	6.478	6.275	6.354			
	Av.	6.261	6.442	6.269	6.376	6.389	6.336±.008	.143±.003
3	441	6.192	6.359	6.287		6.371		
	448	6.219	6.357	6.319	6.342	6.360		
	453	6.253	6.443	6.235	6.340			
	456	6.230	6.514	6.317	6.298			
	457	6.273	6.406	6.297	6.239			
	461	6.232	6.403	6.315		6.375		
	Av.	6.233	6.414	6.295	6.305	6.369	6.317±.009	.166±.003
4	466	6.236	6.327	6.315	6.472	6.314		
	467	6.241	6.290	6.264	6.232	6.331		
	476	6.396	6.443					
	482	6.231		6.350	6.504			
	486	6.242	6.342	6.298	6.387	6.465		
	488		6.324	6.310	6.272			
	Av.	6.269	6.345	6.307	6.373	6.370	6.313±.009	.159±.006
5	501	6.241	6.413	6.357	6.379			
	502	6.247	6.342	6.353	6.273			
	508	6.216	6.291	6.230	6.358			
	514	6.226	6.361	6.217	6.360			
	519		6.400	6.407	6.291	6.302		
	521		6.229	6.300	6.300	6.327		
	Av.	6.232	6.339	6.311	6.327	6.315	6.309±.009	.178±.006
6	530	6.207	6.324	6.407	6.424	6.410		
	541	6.247	6.334	6.301	6.369	6.324		
	544	6.206	6.365	6.276		6.327		
	549	6.236	6.425	6.259	6.336	6.384		
	550	6.220	6.337	6.332	6.415			
	554	6.258	6.306	6.315	6.384	6.392		
	Av.	6.229	6.348	6.315	6.385	6.277	6.318±.008	.168±.006
						Mean	6.320	

Table 12. The pH of the white of fresh eggs.

Pen	Hen	Period 1 2/19-3/2	Period 2 3/8-3/19	Period 3 3/28-4/9	Period 4 4/21-5/4	Period 5 5/13-5/24	Av. pH per pen	Standard Deviation	Correlation coefficient between yolk and white
1	386	8.017	7.982	7.863	8.213	7.836			
	387		7.975	7.758	8.037	7.868			
	391	7.953	7.987	7.826	7.956	7.883			
	392	8.025	7.989	7.781					
	395	7.714	7.968						
	402	7.930	7.903	7.880	7.969	7.609			
	Av.	7.928	7.967	7.821	8.044	7.799	7.935±.014	.214±.010	.36±.056
2	410	7.957	7.989	7.795	7.996				
	411	7.921	7.752	7.582	7.842	7.536			
	421	7.628	7.974	7.738	7.877				
	426	8.024	7.939	7.776	7.922	7.900			
	427		8.044	7.628	7.973	7.893			
	428	8.025	7.949	7.841	7.934				
	Av.	7.911	7.941	7.726	7.924	7.776	7.897±.011	.207±.008	.12±.054
3	441	7.972	7.955	7.754		7.755			
	448	7.912	8.035	7.912	7.998	7.833			
	453	7.869	8.034	7.644	7.810				
	456	7.732	7.943	7.741	7.868				
	457	8.106	8.062	7.888	7.954				
	461	7.918	7.990	7.642		7.745			
	Av.	7.918	8.003	7.763	7.908	7.777	7.894±.013	.233±.009	.41±.045
4	466	7.955	7.883	7.777	7.930	7.810			
	467	7.896	7.971	7.718	7.878	7.894			
	476	8.139	8.071						
	482	7.925		7.896	8.036				
	486	8.122	8.129	7.887	8.235	7.963			
	488		7.950	7.811	7.771				
	Av.	8.007	8.001	7.818	7.970	7.889	7.915±.014	.241±.010	.35±.050
5	501	7.863	8.022	7.860	7.863				
	502	7.990	7.944	7.850	7.813				
	508	8.028	7.948	7.835	8.037				
	514	7.904	8.161	7.836	8.032				
	519		8.091	7.837	8.031	7.943			
	521		7.867	7.710	8.028	7.840			
	Av.	7.946	8.005	7.821	7.967	7.891	7.930±.010	.209±.007	.33±.043
6	530	7.935	8.080	8.043	8.032	7.867			
	541	8.058	8.095	7.686	8.108	7.966			
	514	7.924	7.863	7.834		7.875			
	549	7.880	8.008	7.842	8.123	7.743			
	550	8.058	8.027	7.840	8.056				
	554	7.936	8.028	7.808	8.065	7.912			
	Av.	7.965	8.017	7.842	8.077	7.872	7.950±.010	.211±.007	.28±.043
				Mean	7.920				

Table 13. The averages of the pH of the yolk and white of fresh eggs.*

	Confined. Sunshine thru ordinary glass		Confined. Direct sunshine		Bluegrass range. Direct sunshine	
	pH	Av. pH	pH	Av. pH	pH	Av. pH
White						
No cod liver oil	7.912		7.874		7.925	
With cod liver oil	7.858	7.885	7.937	7.906	7.955	7.940
Yolk						
No cod liver oil	6.317		6.323		6.305	
With cod liver oil	6.347	6.332	6.333	6.328	6.311	6.308

	No cod liver oil		With cod liver oil	
	pH	Av. pH	pH	Av. pH
White				
Confined. Sunshine thru ordinary glass	7.912		7.858	
Confined. Direct sunshine	7.874		7.937	
Open range with bluegrass, sunshine	7.925	7.904	7.955	7.950
Yolk				
Confined. Sunshine thru ordinary glass	6.317		6.347	
Confined. Direct sunshine	6.323		6.333	
Bluegrass range. Direct sunshine	6.305	6.315	6.311	6.330

*The determinations were averaged for each period. The averages were then averaged for each pen and in turn these figures were averaged to obtain the average for each pen.

Table 14. The difference of the means of the pH of yolks of fresh eggs.

Pen	No. obs.	Mean of the pH of egg yolk per pen	Difference	D P. E. diff.
Confined. Sunshine thru ordinary glass				
1 No cod liver oil	114	6.324±.012	.012±.015	0.8
2 With cod liver oil	153	6.336±.008		
Confined. Direct sunshine				
3 No cod liver oil	154	6.317±.009	.004±.013	0.3
4 With cod liver oil	143	6.313±.009		
Bluegrass range. Direct sunshine				
5 No cod liver oil	199	6.309±.009	.009±.012	0.8
6 With cod liver oil	209	6.318±.008		
No cod liver oil				
1 Confined. Sunshine thru ordinary glass	114	6.324±.012	.007±.015	0.5
3 Confined. Direct sunshine	154	6.317±.009		
1 Confined. Sunshine thru ordinary glass	114	6.324±.012	.015±.015	1.0
5 Bluegrass range. Direct sunshine	199	6.309±.009		
3 Confined. Direct sunshine	154	6.317±.009	.008±.012	0.7
5 Bluegrass range. Direct sunshine	199	6.309±.009		
With cod liver oil				
2 Confined. Sunshine thru ordinary glass	153	6.336±.008	.023±.012	1.9
4 Confined. Direct sunshine	143	6.313±.009		
2 Confined. Sunshine thru ordinary glass	153	6.336±.008	.018±.011	1.6
6 Bluegrass range. Direct sunshine	209	6.318±.008		
4 Confined. Direct sunshine	143	6.313±.009	.005±.012	0.4
6 Bluegrass range. Direct sunshine	209	6.318±.008		

Table 15. The difference of the means of the pH of whites of fresh eggs.

Pen	No. obs.	Mean of the pH of egg white per pen	Difference	D P. E. diff.
Confined. Sunshine thru ordinary glass				
1 No cod liver oil	114	7.935±.014		
2 With cod liver oil	153	7.897±.011	.038±.018	2.1
Confined. Direct sunshine				
3 No cod liver oil	154	7.894±.013		
4 With cod liver oil	143	7.915±.014	.021±.019	1.1
Bluegrass range. Direct sunshine				
5 No cod liver oil	199	7.930±.010		
6 With cod liver oil	209	7.950±.010	.020±.014	1.4
No cod liver oil				
1 Confined. Sunshine thru ordinary glass	114	7.935±.014		
3 Confined. Direct sunshine	154	7.894±.013	.041±.019	2.2
1 Confined. Sunshine thru ordinary glass	114	7.935±.014		
5 Bluegrass range. Direct sunshine	199	7.930±.010	.005±.017	0.3
3 Confined. Direct sunshine	154	7.894±.013		
5 Bluegrass range. Direct sunshine	199	7.930±.010	.036±.016	2.3
With cod liver oil				
2 Confined. Sunshine thru ordinary glass	153	7.897±.011		
4 Confined. Direct sunshine	143	7.915±.014	.018±.018	1.0
2 Confined. Sunshine thru ordinary glass	153	7.897±.011		
6 Bluegrass range. Direct sunshine	209	7.950±.010	.053±.015	3.5
4 Confined. Direct sunshine	143	7.915±.014		
6 Bluegrass range. Direct sunshine	209	7.950±.010	.035±.017	2.1

Table 16. The pH of yolk of storage eggs.

Pen	Hen	Group 1	Group 2	Group 3†	Av. pH per pen	Standard Deviation
1	386	7.87	7.71*	8.16*	6.999±.054	.352±.039
	387	-----	6.68*	7.07		
	391	6.61	6.96	7.26		
	392	-----	6.92*	-----		
	395	-----	-----	-----		
	402	7.10*	-----	6.34*		
	Av.	7.19	7.07	7.21		
2	410	7.05	6.98	6.86	6.930±.031	.354±.022
	411	-----	6.19*	-----		
	421	7.36	7.19	7.08		
	426	6.89	7.12	7.03		
	427	6.62	7.08	6.91		
	428	6.89	6.85	6.83		
	Av.	6.96	6.90	6.94		
3	441	6.76	6.89	7.23	7.018±.031	.320±.022
	448	6.89*	6.54*	6.93*		
	453	-----	-----	-----		
	456	7.56	7.22	7.21		
	457	6.98	7.07	7.23		
	461	7.30*	-----	-----		
	Av.	7.10	6.93	7.15		
4	466	6.81	6.81	6.83	7.000±.028	.306±.020
	467	6.98	6.87	6.82		
	476	-----	-----	-----		
	482	-----	7.33	7.14		
	486	6.78*	7.06	7.16*		
	488	7.20	7.38	7.34		
	Av.	6.94	7.09	7.06		
5	501	6.88	7.38	6.95	6.900±.017	.276±.012
	502	6.94	6.96	6.92		
	508	6.91	6.97	7.02		
	514	6.90	6.86	6.93		
	519	6.77	6.93	6.81		
	521	6.71	6.75	6.74		
	Av.	6.85	6.98	6.90		
6	530	-----	7.03*	6.95*	6.910±.026	.334±.019
	541	6.83	6.83	6.99		
	544	6.87	6.84	6.96		
	549	7.04*	7.01	6.92		
	550	7.06	6.82	6.79		
	554	6.96	7.07	6.83		
	Av.	6.95	6.93	6.91		
				Mean	6.976	

*One sample.

†All eggs laid by each hen were divided into three equal lots.

Table 17. The pH of white of storage eggs.

Pen	Hen	Group 1	Group 2	Group 3†	Av. pH per pen	Standard Deviation	Correlation coefficient between yolk and white
1	386	8.61	8.77*	8.72*	8.906±.022	.139±.015	.14±.016
	387	-----	8.98	9.04			
	391	8.92	8.92	9.04			
	392	-----	8.53*	-----			
	395	-----	-----	-----			
	402	8.88*	-----	8.51*			
	Av.	8.80	8.80	8.83			
2	410	9.02	8.59	9.10	8.810±.033	.379±.023	.64±.052
	411	-----	7.64*	-----			
	421	8.63	8.40	8.77			
	426	8.90	8.88	8.82			
	427	8.77	8.85	8.93			
	428	8.83	8.70	9.02			
	Av.	8.83	8.51	8.93			
3	441	8.81	9.02	8.83	8.866±.016	.161±.001	.00±.099
	448	8.61*	8.56*	8.89*			
	453	-----	-----	-----			
	456	8.64	8.82	8.55			
	457	8.88	8.95	8.89			
	461	8.86*	-----	-----			
	Av.	8.76	8.84	8.79			
4	466	8.80	9.03	9.04	8.960±.022	.238±.015	.14±.031
	467	8.78	9.01	8.92			
	476	-----	-----	-----			
	482	-----	8.97	9.00			
	486	9.00*	8.99	8.89*			
	488	8.52	9.08	9.09			
	Av.	8.77	9.02	8.99			
5	501	8.64	8.90	9.02	8.900±.020	.319±.014	.55±.044
	502	8.78	9.01	8.85			
	508	8.85	9.07	9.03			
	514	8.64	8.91	9.01			
	519	8.74	8.97	8.90			
	521	8.65	9.01	8.94			
	Av.	8.72	8.98	8.96			
6	530	-----	8.44*	8.40*	8.930±.017	.215±.012	.70±.041
	541	8.91	8.95	9.00			
	544	8.88	8.96	9.00			
	549	8.90*	9.13	8.96			
	550	8.76	8.94	8.81			
	554	8.86	8.97	9.04			
	Av.	8.86	8.90	8.87			
				Mean	8.895		

*One sample.

†All eggs laid by each hen were divided into three equal lots.

Table 18. The averages of the pH of the yolks and whites of storage eggs.*

	Confined. Sunshine thru ordinary glass		Confined. Direct sunshine		Bluegrass range. Direct sunshine	
	pH	Av. pH	pH	Av. pH	pH	Av. pH
Egg White						
No cod liver oil	8.81		8.80		8.89	
With cod liver oil	8.76	8.79	8.93	8.87	8.88	8.89
Egg Yolk						
No cod liver oil	7.19		7.06		6.91	
With cod liver oil	6.93	7.06	7.03	7.05	6.93	6.92

	No cod liver oil		With cod liver oil	
	pH	Av. pH	pH	Av. pH
Egg White				
1. Confined. Sunshine thru ordinary glass	8.81		8.76	
2. Confined. Direct sunshine	8.80		8.93	
3. Bluegrass range. Direct sunshine	8.89	8.83	8.88	8.86
Egg Yolk				
1. Confined. Sunshine thru ordinary glass	7.19		6.93	
2. Confined. Direct sunshine	7.06		7.03	
3. Bluegrass range. Direct sunshine	6.91	7.05	6.93	6.96

*These figures are the averages of the averages for each hen.

Table 19. The difference of the means of the pH of the yolks of storage eggs.

Pen	No. obs.	Mean of the pH of egg yolk per pen	Difference	D P. E. diff.
Confined. Sunshine thru ordinary glass				
1 No cod liver oil	20	6.999±.054		
2 With cod liver oil	62	6.930±.031	.069±.062	1.1
Confined. Direct sunshine				
3 No cod liver oil	48	7.018±.031		
4 With cod liver oil	55	7.000±.028	.018±.042	0.4
Bluegrass range. Direct sunshine				
5 No cod liver oil	119	6.900±.017		
6 With cod liver oil	75	6.910±.026	.010±.031	0.3
No cod liver oil				
1 Confined. Sunshine thru ordinary glass	20	6.999±.054		
3 Confined. Direct sunshine	48	7.018±.031	.019±.063	0.3
1 Confined. Sunshine thru ordinary glass	20	6.999±.054		
5 Bluegrass range. Direct sunshine	119	6.900±.017	.099±.057	1.7
3 Confined. Direct sunshine	48	7.018±.031		
5 Bluegrass range. Direct sunshine	119	6.900±.017	.118±.036	3.3
With cod liver oil				
2 Confined. Sunshine thru ordinary glass	62	6.930±.031		
4 Confined. Direct sunshine	55	7.000±.028	.070±.042	1.7
2 Confined. Sunshine thru ordinary glass	62	6.930±.031		
6 Buegrass range. Direct sunshine	75	6.910±.026	.020±.040	0.5
4 Confined. Direct sunshine	55	7.000±.028		
6 Buegrass range. Direct sunshine	75	6.910±.026	.090±.038	2.4

Table 20. The difference of the means of the pH of the whites of storage eggs.

Pen	No. obs.	Mean of the pH of egg white per pen	Difference	D P. E. diff.
Confined. Sunshine thru ordinary glass				
1 No cod liver oil	20	8.906±.022		
2 With cod liver oil	62	8.810±.033	.096±.040	2.4
Confined. Direct sunshine				
3 No cod liver oil	48	8.866±.016		
4 With cod liver oil	55	8.960±.022	.094±.027	3.5
Bluegrass range. Direct sunshine				
5 No cod liver oil	119	8.900±.020		
6 With cod liver oil	75	8.930±.017	.030±.026	1.2
No cod liver oil				
1 Confined. Sunshine thru ordinary glass	20	8.906±.022		
3 Confined. Direct sunshine	48	8.866±.016	.040±.027	1.5
1 Confined. Sunshine thru ordinary glass	20	8.906±.022		
5 Bluegrass range. Direct sunshine	119	8.900±.020	.006±.029	0.2
3 Confined. Direct sunshine	48	8.866±.016		
5 Bluegrass range. Direct sunshine	119	8.900±.020	.034±.025	1.4
With cod liver oil				
2 Confined. Sunshine thru ordinary glass	62	8.810±.033		
4 Confined. Direct sunshine	55	8.960±.022	.150±.040	3.8
2 Confined. Sunshine thru ordinary glass	62	8.810±.033		
6 Bluegrass range. Direct sunshine	75	8.930±.017	.120±.037	3.2
4 Confined. Direct sunshine	55	8.960±.022		
6 Bluegrass range. Direct sunshine	75	8.930±.017	.030±.028	1.1

SUMMARY

The pH of the yolk and white of fresh and storage eggs is reported. The eggs were collected from three pairs of pens. The first pair received sunshine thru ordinary window glass. The second were given direct sunshine on a wire-screened porch. The third were given bluegrass range. The hens of one pen of each pair were given cod liver oil. The basal ration was the same for all pens.

Pen averages of fresh eggs showed little variation in pH. The greatest differences occurred between pH 7.894, pen 3, and pH 7.950, pen 6, for the white and pH 6.309, pen 5, and pH 6.336, pen 2, for the yolk. Therefore, it cannot be said that either cod liver oil, sunshine or bluegrass range given to hens, influences the pH of the eggs laid by them.

Storage eggs showed greater variation. The largest differences occurred between pen 2 and pen 4 for white and between pen 5 and pen 3 for yolk, the averages for which were, respectively, 8.810, 8.960, 6.900 and 7.018.

The average pH of yolk and white of fresh eggs was 6.320 and 7.920, respectively. The average pH of yolk and white of storage eggs was 6.976 and 8.895, respectively.

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